

### ABSTRACT OF THE DISCLOSURE

A multilayer enzyme immobilization process is provided comprising adsorbing a polyethyleneimine (PEI) solution in a fibrous matrix, and adding an enzyme to the fibrous matrix, which comprises a plurality of fibrils. The process further comprises forming at least two layers of PEI-enzyme aggregates on the fibrils, and cross-linking the multilayer PEI-enzyme aggregates. The process can further comprise washing the fibrils containing the cross-linked PEI-enzyme aggregates with distilled water and acetic acid buffer subsequent to cross-linking. However, the PEI-containing matrix is not washed prior to the addition of enzyme. The enzyme can be  $\beta$ -galactosidase and the fibrous matrix can be cotton cloth. The multilayer immobilized enzyme can be employed in a biocatalyst reactor for production of galacto-oligosaccharides from lactose and the hydrolysis of lactose to glucose and galactose.